5 steps to live-cell imaging

Follow this guide to capture the best possible images

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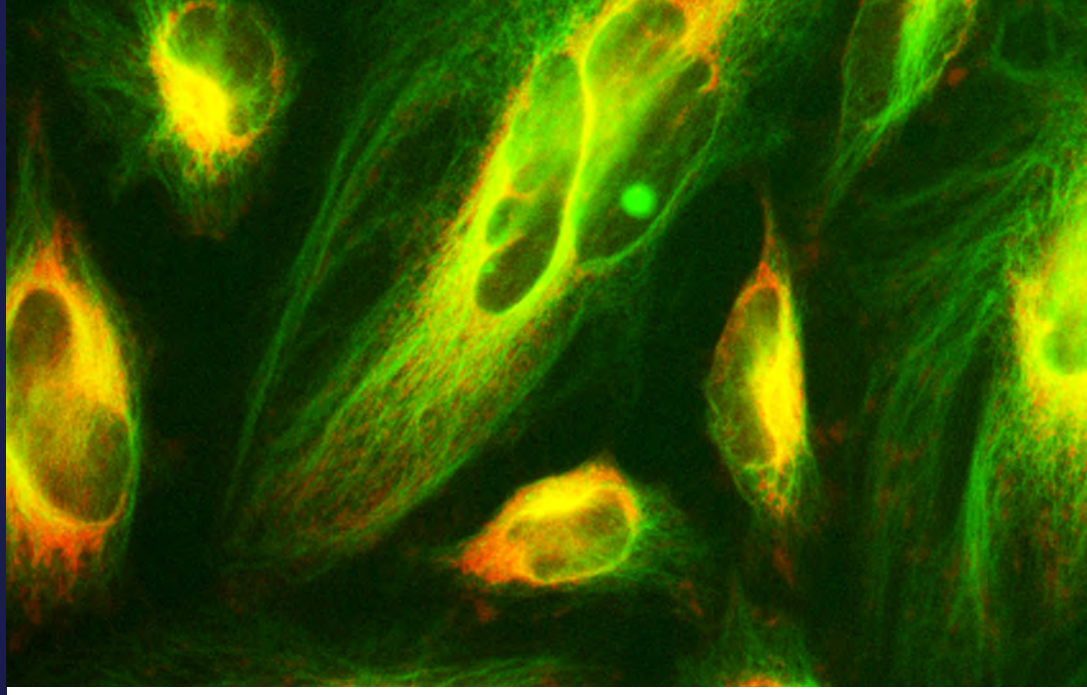
Introduction

Fluorescence imaging of live cells is a powerful approach to the study of dynamic cellular processes and events. Recent advances in fluorescent dye development and innovation have resulted in improved reagents that detect and monitor these dynamic processes. Continued progress in optics, sensor technology, computing power, and software tools have been integrated into imaging systems that are more powerful and straightforward to use.

All of these innovations have contributed to the widespread adoption of fluorescence imaging in cell-based research where scientists apply these new tools in many diverse areas, such as developmental and stem cell biology, medical research, drug discovery, and environmental studies.



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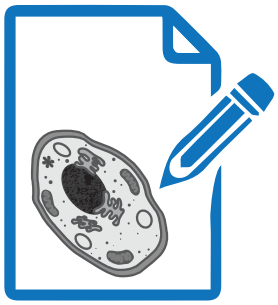

Plan—design your experiment with careful consideration of the tools and resources needed for each step

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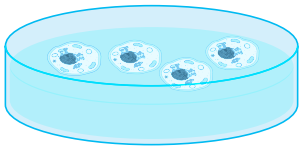
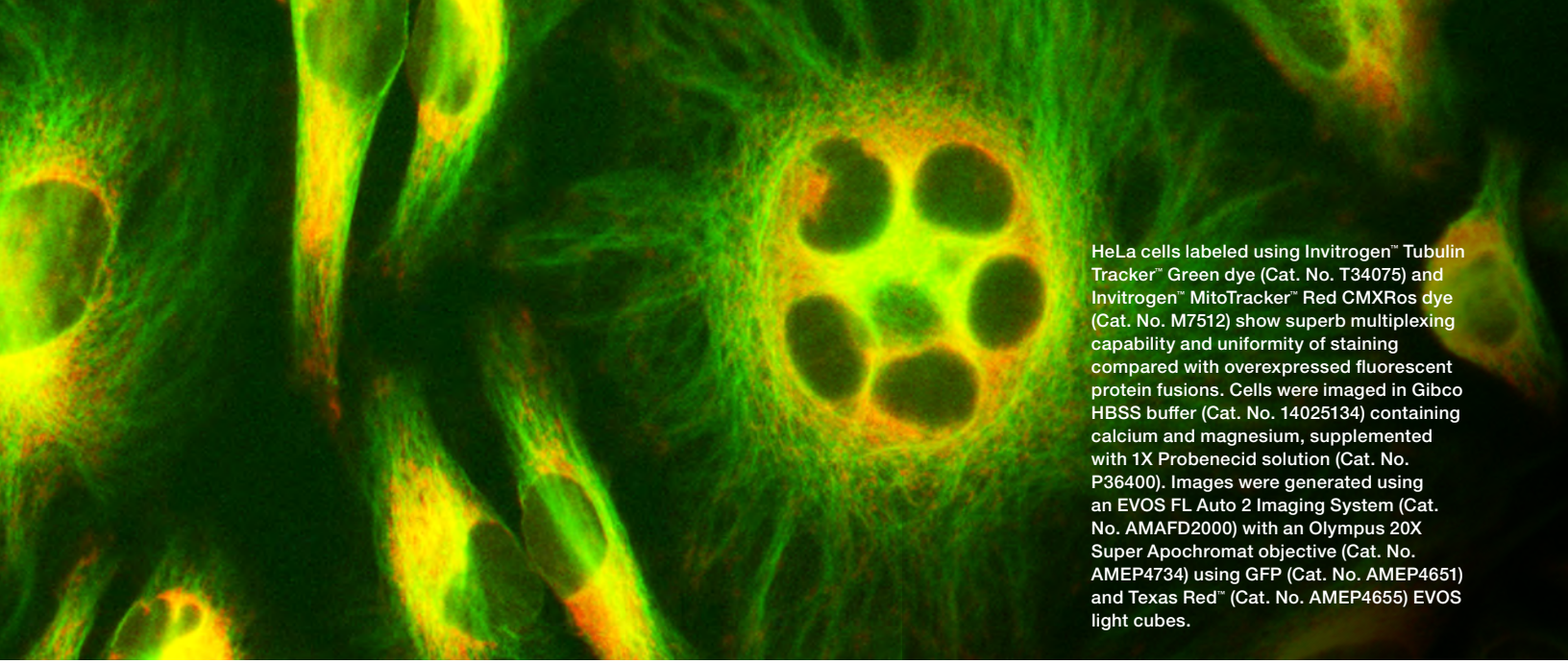


Culture—maintain or grow your cells in optimum conditions

Cover image

Live HeLa cells labeled with Tubulin Tracker™ Deep Red (Cat. No. T34076) and NucBlue™ Live ReadyProbes™ Reagent (Cat. No. R37605), enabling visualization of the microtubule cytoskeleton and nuclei in live cells, respectively. High resolution images were acquired using a confocal system.



HeLa cells labeled using Invitrogen™ Tubulin Tracker™ Green dye (Cat. No. T34075) and Invitrogen™ MitoTracker™ Red CMXRos dye (Cat. No. M7512) show superb multiplexing capability and uniformity of staining compared with overexpressed fluorescent protein fusions. Cells were imaged in Gibco HBSS buffer (Cat. No. 14025134) containing calcium and magnesium, supplemented with 1X Probenecid solution (Cat. No. P36400). Images were generated using an EVOS FL Auto 2 Imaging System (Cat. No. AMAFD2000) with an Olympus 20X Super Apochromat objective (Cat. No. AMEP4734) using GFP (Cat. No. AMEP4651) and Texas Red™ (Cat. No. AMEP4655) EVOS light cubes.

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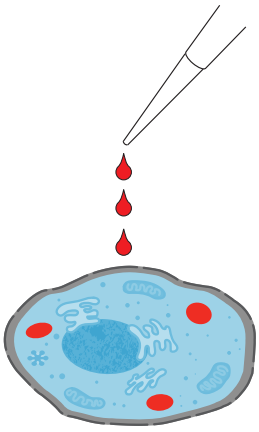

Label—target cell structures, cell functions, and proteins of interest with selective dyes and stains

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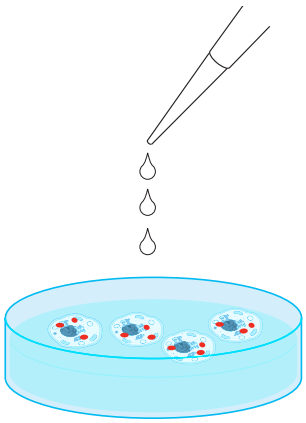

Optimize—minimize background and maintain photostability of fluorescence signals

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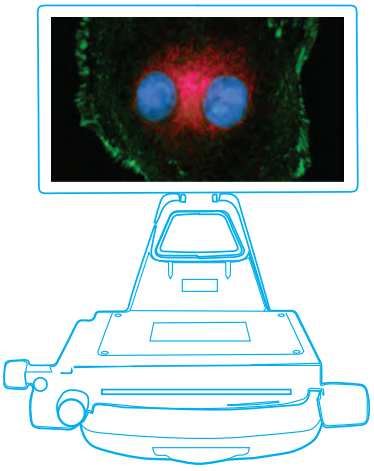


Image—capture discoveries as they happen with maximum clarity and definition

See pages 10–11 for products related to each step.

1 Step 1. Plan

Living cells offer one of the most accessible models of biological processes. Consider the following when deciding whether to use live-cell imaging for your experiment:

Advantages

- Observe dynamic cellular processes as they happen
- Study and image several processes and functions simultaneously using multiplexed assays
- Study cellular structures in their native environment, resulting in more realistic results closer to *in vivo* scenarios
- Track cellular biomolecules and structures over time
- Observe interactions between cells
- Cellular enzymes and other cytosolic biomolecules remain in the cell

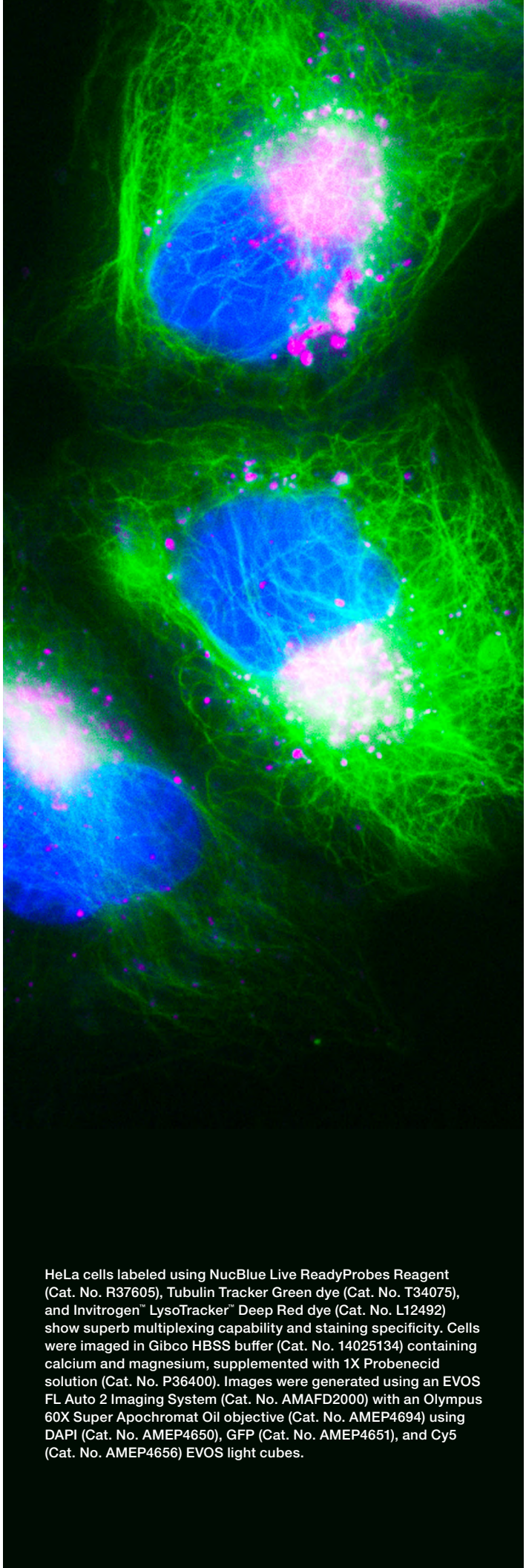
Considerations

- Must have a specific way to label your target with minimal toxicity—whether it is a molecule, a cellular function, or a cellular state
- Living cells are generally not permeable to large detection molecules such as antibodies
- Moving objects can be more difficult to keep in focus
- Certain techniques can be harmful to living cells
- Cells must be kept in their natural physiological state

Resource highlights

Explore **thermofisher.com/5steps-live** where you'll find the following resources and more:

- Reagent selection guide
- Fluorescence SpectraViewer
- Learning center
- Invitrogen™ Molecular Probes™ School of Fluorescence
- Cell staining tool
- Technical support service



HeLa cells labeled using NucBlue Live ReadyProbes Reagent (Cat. No. R37605), Tubulin Tracker Green dye (Cat. No. T34075), and Invitrogen™ LysoTracker™ Deep Red dye (Cat. No. L12492) show superb multiplexing capability and staining specificity. Cells were imaged in Gibco HBSS buffer (Cat. No. 14025134) containing calcium and magnesium, supplemented with 1X Probenecid solution (Cat. No. P36400). Images were generated using an EVOS FL Auto 2 Imaging System (Cat. No. AMAFD2000) with an Olympus 60X Super Apochromat Oil objective (Cat. No. AMEP4694) using DAPI (Cat. No. AMEP4650), GFP (Cat. No. AMEP4651), and Cy5 (Cat. No. AMEP4656) EVOS light cubes.

2 Step 2. Culture

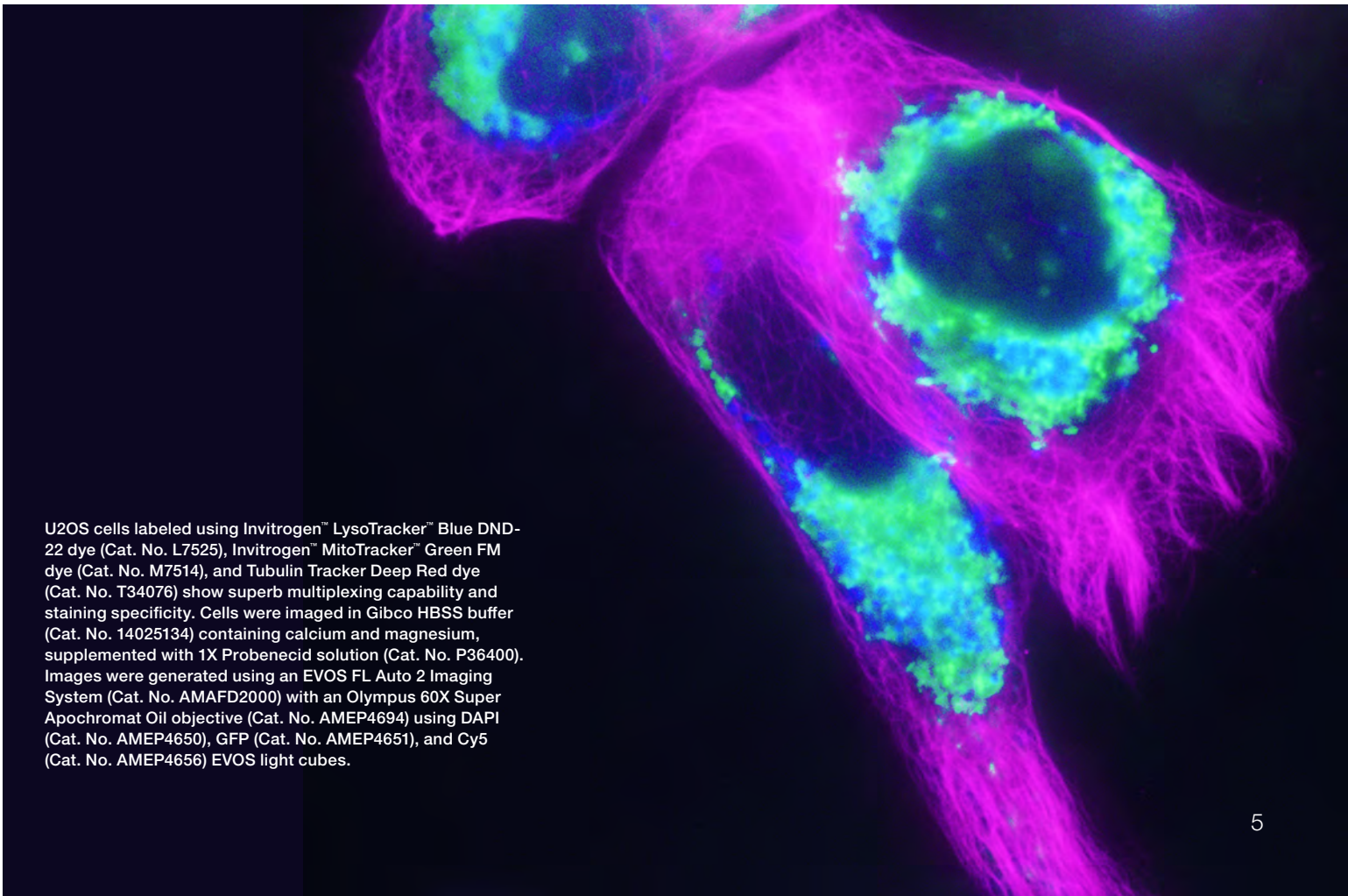
Keeping cells alive and healthy during various experimental manipulations, detection, and imaging is no small task. The choice of medium is particularly important for time-lapse imaging and experiments where cells are exposed to ambient conditions for longer periods. For reliable results with live cells, it is essential that the cells be healthy and kept in an environment as close as possible to physiological temperature, pH, oxygen level, and other conditions.

Tip

You can improve image clarity, reduce background fluorescence, and optimize cell viability by using media and wash buffers created specifically for live-cell imaging and detection (see page 10)

Product highlights

- **Thermo Scientific™ Nunc™ cell culture vessels with Nunclon™ Delta surface treatment** have been validated with Gibco™ media to confirm consistent cell growth across multiple cell lines. It's a proven combination for happy cells and happy scientists. Find out more at **thermofisher.com/cellcultureplastics**
- **Gibco™ extracellular matrices, scaffolds, and proteins** provide *in vivo*–like morphology and physiologically relevant environments for more realistic cell biology and better intercellular interactions. Find out more at **thermofisher.com/3dcellculture**
- **The Invitrogen™ Countess™ II FL Automated Cell Counter** is an affordable and automated tool for checking the health of your cells quickly and objectively. It has reusable or disposable slide options. Find out more at **thermofisher.com/countess**



U2OS cells labeled using Invitrogen™ LysoTracker™ Blue DND-22 dye (Cat. No. L7525), Invitrogen™ MitoTracker™ Green FM dye (Cat. No. M7514), and Tubulin Tracker Deep Red dye (Cat. No. T34076) show superb multiplexing capability and staining specificity. Cells were imaged in Gibco HBSS buffer (Cat. No. 14025134) containing calcium and magnesium, supplemented with 1X Probenecid solution (Cat. No. P36400). Images were generated using an EVOS FL Auto 2 Imaging System (Cat. No. AMAFD2000) with an Olympus 60X Super Apochromat Oil objective (Cat. No. AMEP4694) using DAPI (Cat. No. AMEP4650), GFP (Cat. No. AMEP4651), and Cy5 (Cat. No. AMEP4656) EVOS light cubes.

3 Step 3. Label

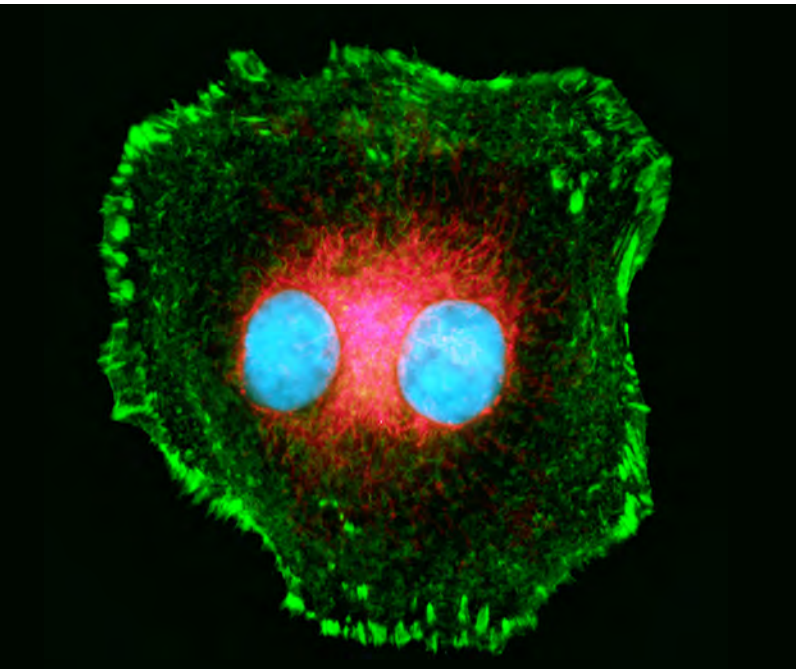
The appropriate fluorophore (targeted fluorescent protein or small membrane-permeant reagent) should be used to monitor your target cellular structure or process. Additional fluorophores can be used to monitor multiple cellular structures and processes, but the excitation and emission spectra should be checked using the Fluorescence SpectraViewer to ensure minimal spectra overlap.

It is critical to avoid using too much fluorescent label because excessive fluorescent labeling can result in:

- Nonspecific staining with increased background signals
- Physiological artifacts and structural perturbations
- Cytotoxicity
- Spectral overlap

Note:

- **Live-cell structure reagents** help identify cellular components
- **Live-cell function reagents** help identify cellular functions and processes



HeLa cells were transduced with CellLight Mitochondria-RFP reagent (Cat. No. C10505) and CellLight Talin-GFP reagent (Cat. No. C10611) for 24 hours, then labeled with NucBlue Live ReadyProbes Reagent (Cat. No. R37605) for 15 minutes. For photobleach protection, cells were incubated with ProLong Live Antifade Reagent (thermofisher.com/prolonglive) for 90 minutes before imaging on an EVOS cell imaging system (thermofisher.com/evos).

Tips

- Consider using a longer-wavelength fluorescent reagent if extended light exposure is required. This will require lower excitation power, which can correlate to lower phototoxicity and healthier cells.
- Staining must be optimized for the particular assay readout, spectral compatibility, and signal-to-background ratio.
- Removing the labeling solution and rinsing with fresh medium will reduce background fluorescence.

Product highlights

Invitrogen™ CellLight™ reagents have proven to be the easiest to use for labeling specific structures in live cells. Targeted fluorescent proteins are introduced using the Invitrogen™ BacMam™ transduction system; no molecular biology techniques are required. Simply add the reagent to your cells, incubate overnight, and you're ready to image in the morning. Get more information at **thermofisher.com/celllight**

Invitrogen™ CellTracker™ reagents are a diverse reagent class used for labeling mammalian cells to view changes in morphology or location. These nontoxic fluorescent dyes are designed to freely pass through cell membranes into cells, where they are transformed into cell-impermeant reaction products. Incubating cells with a CellTracker reagent for 30 minutes will provide at least 72 hours of fluorescent signal (typically three to six generations). Get more information at **thermofisher.com/celltracking**

Invitrogen™ pHrodo™ indicators are fluorogenic dyes that dramatically increase in fluorescence as the pH of their surroundings becomes more acidic. When conjugated to dextrans, proteins, or other particles, pHrodo dyes can be used as highly specific sensors of endocytic and phagocytic internalization and lysosomal sequestration in live cells, offering a superior alternative to conjugates of other fluorescent dyes such as fluorescein and tetramethylrhodamine. Get more information at **thermofisher.com/phrodo**

See the comprehensive product list on pages 10–11 or at **thermofisher.com/5steps-live**

4 Step 4. Optimize

Signal-to-background ratio can be optimized by using reagents that reduce extracellular fluorescence and increase fluorophore photostability. It is important to image in media that have been specifically designed for maintaining cell health while reducing or eliminating background fluorescence in live-cell imaging experiments (see Table 1). The addition of a background suppressor compatible with live cells can also help reduce extracellular background fluorescence and eliminate the need for a wash step. Antifade mounting media for live cells can be applied to samples to reduce photobleaching of fluorophores, preventing signal loss with multiple or long exposures.

Table 1. Step 4 product comparison.

Reagent	Cell washing	Short-term imaging	Imaging up to 4 hours	Long-term imaging
Gibco™ PBS, pH 7.4	✓	✓		
Invitrogen™ Live Cell Imaging Solution	✓	✓	✓	
Gibco™ FluoroBrite™ DMEM	✓	✓	✓	✓

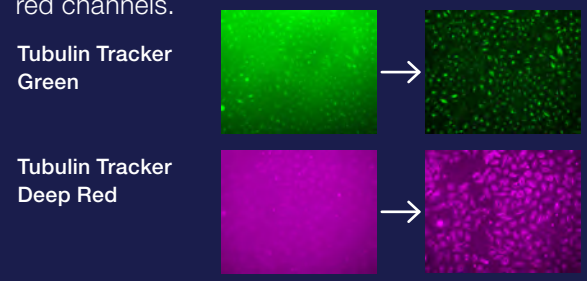
Tips

- If no further culture is planned, a background suppressor can be used to optimize the signal by reducing the haze and increasing the contrast.
- The use of an antifade reagent has been shown to increase fluorophore photostability and decrease the effect of phototoxicity in a variety of sample types.

- **Gibco PBS, pH 7.4** (Cat. No. 10010023) is ideal for cell washing and short-term imaging where prolonged incubation is not required.
- **Invitrogen Live Cell Imaging Solution** (Cat. No. A14291DJ) is an optically clear solution used for imaging, dye loading, and wash steps. It helps keep cells healthy for up to 4 hours.

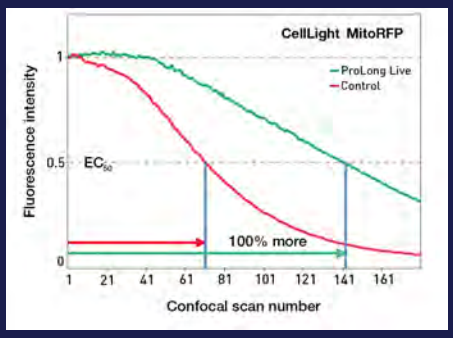
Product highlights

- **Invitrogen™ BackDrop™ Background Suppressor** (Cat. No. B10512) is used when observing high background signal or weak fluorescence in the blue, green, or red channels.

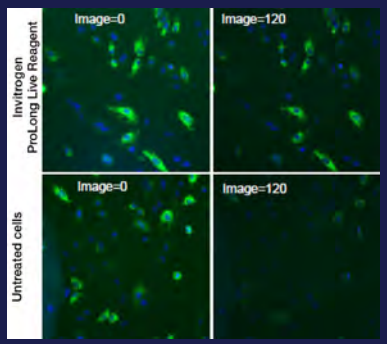


Live HeLa cells labeled with Tubulin Tracker Green dye (Cat. No. T34075) and Tubulin Tracker Deep Red dye (Cat. No. T34076). Addition of BackDrop Background Suppressor greatly reduces extracellular background while leaving intracellular labeling unaffected **(right)**.

- **Invitrogen™ ProLong™ Live Antifade Reagent** (Cat. No. P36974, P36975) is used to increase fluorophore photostability. See the difference at **thermofisher.com/prolonglive**



The overall signal protection offered by ProLong Live reagent compared to untreated samples is calculated based on the scan number where treated and untreated samples reach the EC_{50} value. The addition of ProLong Live reagent permitted 100% more captures with CellLight Mitochondria-RFP reagent.



After 120 exposures using a standard time-lapse imaging protocol, samples treated with ProLong Live reagent are >20% brighter than untreated cells, enabling more data collection time.

5 Step 5. Image

Live-cell imaging of dynamic processes requires active observation over time.

Illumination and detection

To minimize phototoxicity, choose imaging systems that give you the greatest control of light sources. Try to minimize light intensity, exposure time, wavelength range, and amount of excitation energy for illuminating your cells while still generating a good signal with low background. Use the illumination that gives you the highest signal with the lowest level of fluorophore excitation. In some cases (particularly when you wish to image over a long period of time), it is advisable to sacrifice resolution by using shorter exposure times or lower magnification in exchange for healthier cells.

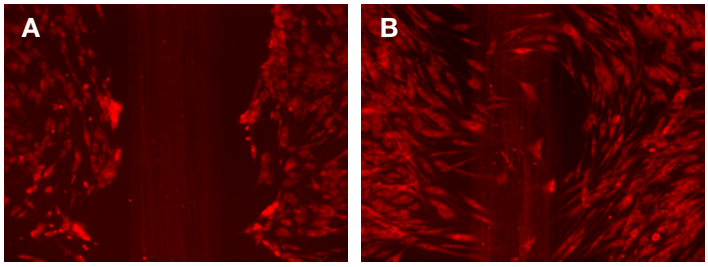
Live-cell imaging over longer periods of time can be challenging because the target may move out of focus during the course of the experiment. Many microscopes have autofocusing features that can help keep your target in focus longer and reduce focal drift. Additionally, maintaining cells at a constant temperature and keeping the volume of solution in the vessel constant will help with focal drift.

Environmental control

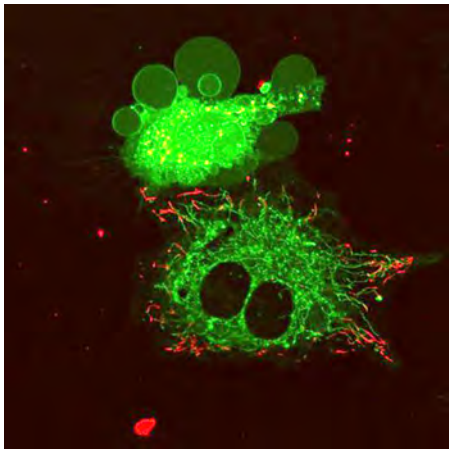
Many cells cannot tolerate deviations from their optimal temperature, osmolarity, pH, and humidity. Requirements vary depending on what experimental question you are asking. For example, experiments investigating cell growth and division may have a different set of requirements than experiments involving receptor activation and calcium accumulation. Some robust immortalized cell lines will tolerate being imaged or monitored for short periods of time without any environmental control. Conversely, for long-term imaging and detection studies, good results with both immortalized cells and primary cells typically require tightly controlled environmental parameters.

Tips

- For short-term imaging experiments, use a large volume of imaging medium to prevent changes in osmolarity and oxygen resulting from evaporation of the medium.
- To focus on a sample, start with a low magnification. This will minimize the time the sample is exposed to light.
- Avoid using autofocus for every image taken during time-lapse imaging. Autofocus can increase the amount of light energy hitting the sample by as much as 10 times.
- For longer time-course imaging or imaging of sensitive cells, an onstage incubator may be added to the imaging equipment to allow precious control of temperature, humidity, and CO_2 levels.



A scratch wound in a culture of HDFn cells loaded with Invitrogen™ CellTracker™ Deep Red Dye (Cat. No. C34565). (A) The illuminated area was subjected to repeated illumination for 10 hours. Cells in this area show signs of phototoxicity (a loss of viability as cells were not able to grow into the wound). **(B)** Cells in the non-illuminated area show viable cell growth into the wound.



The top cell shows catastrophic blebbing of the cell membrane caused by excessive light exposure. Blebbing is a term used to describe membrane perturbation caused by toxicity. By contrast, the bottom cell remains relatively healthy and is not displaying aberrant morphology.

Product highlights

Cell counter

- To avoid the pitfall of proceeding to the next step in your experiment with unhealthy cells, a quick check for cell health can be done on the **Countess II FL Automated Cell Counter** when used in conjunction with a variety of fluorescent reagents to detect cell viability, apoptosis, cytotoxicity, and transfection efficiency. The reusable slide option reduces consumption cost. Find out more at **thermofisher.com/countess**

Countess II FL Automated Cell Counter

Microscopy

- Designed specifically for **Invitrogen™ EVOS™ imaging systems**, the **Invitrogen™ EVOS™ Onstage Incubator** is an environmental chamber that enables precise control of temperature, humidity, and three gases for time-lapse imaging of live cells under both physiological and nonphysiological conditions. Find out more at **thermofisher.com/evos-osi**

Invitrogen™ EVOS™ M5000 Cell Imaging System with EVOS Onstage Incubator

High-content analysis (HCA)

- **The Invitrogen™ HCA Onstage Incubator** for Thermo Scientific™ CellInsight™ HCA platforms allows precise control of temperature, humidity, and CO_2 levels so that you may observe and measure biological activity and changes over time. Data gathered from longer-term imaging studies are the basis of quantitative analysis studies, especially when combined with Thermo Scientific™ HCS Studio™ Software for increased statistical power. Find out more at **thermofisher.com/hcaosi**

Thermo Scientific™ CellInsight™ CX7 LZR High-Content Screening Platform with HCA Onstage Incubator

Assess cell health before imaging

Invitrogen™ cell health assays have shown excellent results on the Thermo Scientific™ Varioskan™ LUX Multimode Microplate Reader equipped with gas module, which can read a 96-well plate in as little as six seconds. Find out more at **thermofisher.com/varioskanlux**

Varioskan LUX Multimode Microplate Reader

Selection guide for live-cell imaging

Use the table below to find the tools you need for each step

Step 1. Plan	Planning tools	Fluorescence SpectraViewer (**thermofisher.com/spectraviewer**) Cell Staining Tool (**thermofisher.com/cellstaintool**) Cell Analysis Learning Center (**thermofisher.com/celllearning**) Cell Analysis Support Center (**thermofisher.com/cellsupport**) Contact us (**thermofisher.com/contact**)	
Step 2. Culture	Culturewares, media, buffers	Nunc chamber slide system, coverglass, and glass-bottom dishes (**thermofisher.com/cellculturesplastics**) CoverWell Perfusion Chamber Gasket (C18139) FluoroBrite DMEM (**thermofisher.com/fluorobrite**, A1896702) B-27 Plus Neuronal Culture System (**thermofisher.com/b27plus**, A3653401) Fetal bovine serum (**thermofisher.com/fbs**) GlutaMAX Supplement (35050061)	
	Extracellular matrices	Geltrex matrix (A1569601) Poly-D-Lysine (A3890401) Laminin (23017015)	
	Transfection	Invitrogen transfection reagents (**thermofisher.com/transfection**)	
	Growth factors	Gibco growth factors (**thermofisher.com/growthfactors**)	
Step 3. Label	**Cell structure**	**Blue**	**Green**
	Plasma membrane	CellLight Plasma Membrane-CFP (C10606)	CellMask Green Plasma Membrane (C37608) CellLight Plasma Membrane-GFP (C10607)
	Nucleus	NucBlue Live ReadyProbes Reagent (R37605) CellLight Nucleus-CFP (C10616)	SYTO 9 Green Nucleic Acid (S34854) CellLight Nucleus-GFP (C10602)
	Cytoskeleton	Alexa Fluor Plus 405 Phalloidin (A30104)	Tubulin Tracker Green (T34075) CellLight Actin-GFP (C10582) CellLight Tubulin-GFP (C10509)
	Endoplasmic reticulum		ER-Tracker Green (E34251) CellLight ER-GFP (C10590)
	Lysosomes		LysoTracker Green (L7526) CellLight Lysosomes-GFP (C10596)
	Mitochondria		MitoTracker Green FM (M7514) CellLight Mitochondria-GFP (C10508)
	Cell tracking	CellTracker Blue CMF_2HC (C12881) CellTracker Violet BMQC (C10094) CellTracker Blue CMAC (C2110)	CellTracker Green CMFDA (C7025) Qtracker 525 Cell Labeling Kit (Q25041MP)
	Cell function	**Blue**	**Green**
	Viability	ReadyProbes Cell Viability Imaging Kit, Blue/Green (R37609) ReadyProbes Cell Viability Imaging Kit, Blue/Red (R37610) Calcein Blue, AM (C1429)	LIVE/DEAD Viability/Cytotoxicity Kit (L3224) ReadyProbes Cell Viability Imaging Kit, Blue/Green (R37609) LIVE/DEAD Cell Imaging Kit (R37601) Calcein, AM (C3100MP)
	Oxidative stress detection	ThiolTracker Violet (T10095)	CellROX Green (C10444) Click-iT Lipid Peroxidation Imaging Kit (C10446) Premo Cellular Hydrogen Peroxide Sensor (P36243) Premo Cellular Redox Sensor Grx-1-roGFP (P36242)
	Apoptosis (Ap) and autophagy (Au)		CellEvent Caspase-3/7 Green (Ap) (C10423) Premo Autophagy Sensor LC3B-GFP (Au) (P36235) Premo Autophagy Tandem Sensor RFP-GFP-LC3B (Au) (P36239)
	Endocytosis (E) and phagocytosis (P)		pHrodo Green Zymosan Bioparticles Conjugate (P) (P35365) pHrodo Green *E. coli* Bioparticles Conjugate (P) (P35366) pHrodo Green *S. aureus* Bioparticles Conjugate (P) (P35367) CellLight Early Endosomes-GFP (C10586) CellLight Late Endosomes-GFP (C10588) pHrodo Green Dextran, 10,000 MW (E) (P35368) Transferrin from Human Serum, Alexa Fluor 488 Conjugate (E) (T13342) pHrodo iFL Green Microscale Protein Labeling (P/E) (P36015) Zenon pHrodo iFL Green Human IgG Labeling (E) (Z25611) Zenon pHrodo iFL Green Mouse IgG Labeling (E) (Z25609)
	Antibody internalization		pHrodo iFL Green Microscale Protein Labeling Kit (P36015) Zenon pHrodo iFL Green Human IgG Labeling Reagent (Z25611) Zenon pHrodo iFL Green Mouse IgG Labeling Reagent (Z25609)
	Proliferation		Click-iT Plus EdU Alexa Fluor 488 Imaging Kit (C10637) Click-iT EdU Alexa Fluor 488 Imaging Kit (C10337)
	Ion (I) and membrane (M) potential indicators	SBFI Sodium Ions (I) (S1263)	Fluo-4, AM Calcium Ions (I) (F14201) Fluo-4 Calcium Imaging Kit (I) (F10489) FluoVolt Membrane Potential Kit (M) (F10488) FluxOR II Potassium Ion Channel (I) (F20015) FluoZin-1, AM Zinc Ions (I) (F24180) FluoZin-3, AM Zinc Ions (I) (F24194) CoroNa Green Sodium Ions (I) (C36676) Magnesium Green, AM (I) (M3735)
Step 4. Optimize	Media and solutions	PBS, pH 7.4 (10010023) Live Cell Imaging Solution (A14291DJ) FluoroBrite DMEM (**thermofisher.com/fluorobrite**)	
	Background suppressor	BackDrop Background Suppressor (B10512)	
	Mountant and antifade reagents	ProLong Live antifade reagents (**thermofisher.com/prolonglive**)	
Step 5. Image	Imaging and analysis reagents	Countess cell counters (**thermofisher.com/countess**) EVOS imaging systems with onstage incubator (**thermofisher.com/evos**) CellInsight high-content analysis platforms with onstage incubator (**thermofisher.com/hca**)	

Find out more at **thermofisher.com/5steps-live**

Working together to analyze your cells

Using the right combination of analysis platforms helps enable experiments that would otherwise be limited by a competing need for instrumentation in your lab. When you perform time-lapse imaging using the EVOS FL Auto 2 Imaging System with the EVOS Onstage Incubator, even a multiday imaging experiment will not interfere with your ability to measure calcium flux or gene expression using luminescence on the Varioskan LUX multimode reader.

Orange	Red	Deep Red
CellMask Orange Plasma Membrane (C10045) CellLight Plasma Membrane-RFP (C10608)		CellMask Deep Red Plasma Membrane (C10046)
SYTO 82 Orange Nucleic Acid (S11363) CellLight Nucleus-RFP (C10603)	SYTO 59 Red Nucleic Acid (S11341)	NucRed Live 647 ReadyProbes (R37106)
CellLight Actin-RFP (C10502) CellLight Tubulin-RFP (C10503)		Tubulin Tracker Deep Red (T34076)
CellLight ER-RFP (C10591)	ER-Tracker Red (E34250)	
LysoTracker Red (L7528) CellLight Lysosomes-RFP (C10597)		LysoTracker Deep Red (L12492)
MitoTracker Orange CMTMRos (M7510) CellLight Mitochondria-RFP (C10505)	MitoTracker Red CM-H_2Xros (M7513)	MitoTracker Deep Red FM (M22426)
CellTracker Orange CMRA (C34551) Qtracker 585 Cell Labeling Kit (Q25011MP)	CellTracker Red CMTPX (C34552) Qtracker 605 Cell Labeling Kit (Q25001MP)	CellTracker Deep Red (C34565) Qtracker 655 Cell Labeling Kit (Q25021MP)
Orange	**Red**	**Deep Red**
ReadyProbes Cell Viability Imaging Kit, Blue/Red (R37610)	LIVE/DEAD Viability/Cytotoxicity Kit (L3224) LIVE/DEAD Cell Imaging Kit (R37601)	
CellROX Orange (C10443)	MitoSOX Red Mitochondrial Superoxide (M36008)	CellROX Deep Red (C10422)
Premo Autophagy Sensor LC3B-RFP (Au) (P36236) Premo Autophagy Tandem Sensor (P36239) RFP-GFP-LC3B (Au) (P36239)		
pHrodo Red Zymosan Bioparticles Conjugate (P) (P35364) pHrodo Red *E. coli* Bioparticles Conjugate (P) (P35361) pHrodo Red *S. aureus* Bioparticles Conjugate (P) (A10010) CellLight Early Endosomes-RFP (C10587) CellLight Late Endosomes-RFP (C10589) pHrodo Red Dextran, 10,000 MW (E) (P10361) pHrodo Red Transferrin Conjugate (E) (P35376) Transferrin from Human Serum, Alexa Fluor 555 Conjugate (E) (T35352) pHrodo Red Epidermal Growth Factor Conjugate (E) (P35374) pHrodo iFL Red Microscale Protein Labeling (P/E) (P36014) Zenon pHrodo iFL Red Mouse IgG Labeling (E) (Z25610) Zenon pHrodo iFL Red Human IgG Labeling (E) (Z25612)	Transferrin from Human Serum, Alexa Fluor 594 Conjugate (E) (T13342)	Transferrin from Human Serum, Alexa Fluor 647 Conjugate (E) (T23366)
pHrodo Red Epidermal Growth Factor Conjugate (P35374) pHrodo iFL Red Microscale Protein Labeling Kit (P36014) SiteClick Antibody Azido Modification Kit (S20026) Click-iT pHrodo iFL Red sDIBO Alkyne for Antibody Labeling (C20034) Zenon pHrodo iFL Red Human IgG Labeling Reagent (Z25612) Zenon pHrodo iFL Red Mouse IgG Labeling Reagent (Z25610)	pHrodo iFL Red Microscale Protein Labeling Kit (P36014)	
Click-iT Plus EdU Alexa Fluor 555 Imaging Kit (C10638) Click-iT EdU Alexa Fluor 555 Imaging Kit (C10338)	Click-iT Plus EdU Alexa Fluor 594 Imaging Kit (C10639) Click-iT EdU Alexa Fluor 594 Imaging Kit (C10339)	Click-iT Plus EdU Alexa Fluor 647 Imaging Kit (C10640) Click-iT EdU Alexa Fluor 647 Imaging Kit (C10340)
Rhod-2, AM Calcium Ions (I) (R1245MP) Rhod-3 Calcium Imaging Kit (I) (R10145)	FluxOR Red Potassium Ion Channel (I) (F20018)	

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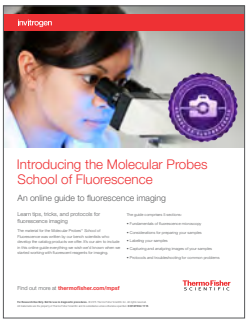

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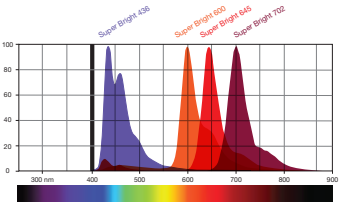

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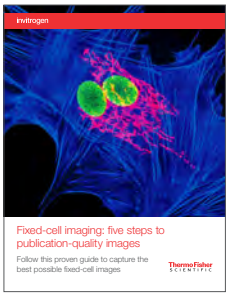

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